

Analysis of Plant Tissues Using Vibrational and Other Spectroscopic Methods and Multivariate Approaches

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Understanding the structure and function of plant tissues requires the combination of analytical tools that govern different scales, ranging from the micro-morphology to the molecular composition, and that elucidate the interaction of organic and inorganic materials. While Raman spectroscopy as a vibrational method allows to investigate the structure and chemical composition of plant tissue sections, second harmonic generation (SHG) and fluorescence microscopy give complementary insight into the orientation, morphology, and heterogeneity of the distribution of the tissue building blocks at the microscopic level. Here, Raman microscopy was combined with SHG, two-photon excited fluorescence (2PF), [1] and scanning electron microscopy (SEM-EDX) to investigate several different plant tissues including e.g., pollen grains and cells walls in plant sections, and to image their composition and structure. Using multivariate techniques, specifically hierarchical cluster analysis (HCA) and principal component analysis (PCA) on the Raman data sets, histological characterization of these tissues is possible. In pollen tissues, differentiation of histological substructures (pollen grain center, pollen tube shank or apex) is possible using PCA, and is an important prerequisite in order to identify small biological differences brought about by altered physiological situations [2]. Similarly, we present an approach to refold subspaces of PCA scores plots so that a visual identification of physiological differences is possible from multivariate tissue maps. As an example, we discuss the influence of silica both in the germination medium in pollen grains, and during the growth of whole plants. Silica also plays an important role in plants, as it can form microscopic silica deposits, called phytoliths. Variations in the silica nanostructure between the different phytolith cell types were observed using both, Raman techniques and SEM-EDX. We conclude that only the combination of all these different analytical tools can give a full picture of the influence of specific growth conditions on the structure of complex plant tissues.

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References

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